

We claim:

1. A method of determining a true signal of an analyte, comprising:

(a) measuring an observed signal x for one or 5 more analytes, and

(b) determining a mean signal (μ) and a system parameter (β) for said analyte that produce enhanced values for a probability likelihood of said observed signal, said observed signal being related to said mean 10 signal by an additive error (δ) and a multiplicative error (ε), wherein said system parameter specifies properties of said additive error (δ) and said multiplicative error (ε).

2. The method of claim 1, further comprising 15 selecting a mean signal μ that provides a maximum probability of likelihood given said observed signal.

3. The method of claim 1, wherein said additive and multiplicative errors are independent with respect to each other.

20 4. The method of claim 1, wherein said observed signal and said mean signal further comprises the relationship:

$$x_{ij} = \mu_{xi} + \mu_{xi}\varepsilon_{xij} + \delta_{xij},$$

where each measurement $j=1, \dots, M$, each analyte $i=1, 25 \dots, N$, and where x_{ij} is the observed signal and μ_{xi} is the mean signal.

5. The method of claim 1, wherein said additive and multiplicative errors further comprise a univariate distribution.

6. The method of claim 5, wherein said 5 univariate distribution is a parametric distribution.

7. The method of claim 6, wherein said parametric distribution is a univariate normal distribution.

8. The method of claim 7, wherein said 10 univariate normal distribution and said system parameter further comprise a multiplicative error term consisting of a normal distribution having standard deviation with respect to a signal mean (σ_{ex}), and an additive error term consisting of a normal distribution having standard 15 deviation with respect to a signal mean ($\sigma_{\delta x}$).

9. The method of claim 6, wherein said parametric distribution is a t-distribution.

10. The method of claim 6, wherein said parametric distribution is a gamma distribution.

20 11. The method of claim 1, wherein said mean signal and system parameter are determined at the same time.

12. The method of claim 1, wherein said system parameter is determined before said mean signal is 25 determined.

13. The method of claim 12, wherein said predetermined system parameter is used to determine said mean signal.

14. The method of claim 1, wherein said 5 enhanced values for said probability likelihood of said observed signals are produced one or more times until said mean signal and said system parameter converge.

15. The method of claim 1, wherein said mean signal and said system parameter are determined by a 10 method selected from the group consisting of maximum likelihood estimation (MLE), Quasi-Maximum Likelihood and Generalized Method of Moments.

16. The method of claim 1, wherein determining said mean signal and said system parameter further 15 comprises a non-linear optimization algorithm.

17. The method of claim 16, wherein said optimization algorithm is selected from the group consisting of Gradient Descent, Newton-Raphson and Simulated Annealing.

18. A method of determining a true signal of an analyte, comprising:

- (a) obtaining an observed signal x for one or more analytes;
- 5 (b) providing a mean signal (μ) and a system parameter (β) for said analyte;
- (c) computing a probability likelihood of said observed signal, said observed signal being related to said mean signal by an additive error (δ) and a
- 10 multiplicative error (ε), where said system parameter specifies properties of said additive error and said multiplicative error, and
- (d) selecting a mean signal μ and a system parameter (β) that provides a maximum probability
- 15 likelihood of occurrence given said observed signal.

19. The method of claim 18, wherein said additive and multiplicative errors are independent with respect to each other.

20. The method of claim 18, wherein said observed signal and said mean signal further comprises the relationship:

$$x_{ij} = \mu_{xi} + \mu_{xi}\varepsilon_{xij} + \delta_{xij},$$

where each measurement $j=1, \dots, M$, each analyte $i=1, \dots, N$, and where x_{ij} is the observed signal and μ_{xi} is the

25 mean signal.

21. The method of claim 18, wherein said additive and multiplicative errors further comprise a univariate distribution.

22. The method of claim 1, wherein said univariate distribution is a parametric distribution.

23. The method of claim 22, wherein said parametric distribution is a univariate normal
5 distribution.

24. The method of claim 23, wherein said univariate normal distribution and said system parameter further comprise a multiplicative error term consisting of a normal distribution having standard deviation with
10 respect to a signal mean (σ_{ex}), and an additive error term consisting of a normal distribution having standard deviation with respect to a signal mean ($\sigma_{\delta x}$).

25. The method of claim 22, wherein said parametric distribution is a t-distribution.

15 26. The method of claim 22, wherein said parametric distribution is a gamma distribution.

27. The method of claim 18, wherein said mean signal and system parameter are selected at the same
20 time.

28. The method of claim 18, wherein said system parameter is selected before said mean signal is determined.

29. The method of claim 28, wherein said
25 preselected system parameter is used to select said mean signal.

30. The method of claim 18, further comprising computing said probability likelihood one or more times until said mean signal and said system parameter converge.

5 31. The method of claim 18, wherein said mean signal and said system parameter are determined by a method selected from the group consisting of maximum likelihood estimation (MLE), Quasi-Maximum Likelihood and Generalized Method of Moments.

10 32. The method of claim 18, wherein selecting said mean signal and said system parameter further comprises a non-linear optimization algorithm.

15 33. The method of claim 32, wherein said optimization algorithm is selected from the group consisting of Gradient Descent, Newton-Raphson and Simulated Annealing.

34. A method of determining relative amounts of an analyte between samples, comprising:

20 (a) measuring observed signals x and y for an analyte within two or more sample pairs, and
(b) determining a mean signal pair per analyte (μ) and a system parameter (β) for each sample pair that produce enhanced values for a probability likelihood of said observed signals, said observed signals being
25 related to said mean signals by an additive error (δ) and a multiplicative error (ε), wherein said system parameter specifies properties of said additive error (δ) and said multiplicative error (ε).

35. The method of claim 34, further comprising selecting a mean signal μ that provides a maximum probability of occurrence given said observed signals.

5 36. The method of claim 34, wherein said additive and multiplicative errors are independent with respect to each other.

37. The method of claim 34, wherein said observed signals and said mean signal pair per analyte 10 within said sample pairs further comprise the relationship:

$$x_{ij} = \mu_{xi} + \mu_{xi}\varepsilon_{xij} + \delta_{xij}, \text{ and}$$
$$y_{ij} = \mu_{yi} + \mu_{yi}\varepsilon_{yij} + \delta_{yij}$$

15 where each measurement j equals 1 through M and each analyte i equals 1 through N ; where x_{ij} and y_{ij} are the observed signals, and where μ_{xi} and μ_{yi} are the mean signals.

20 38. The method of claim 34, wherein said additive and multiplicative errors further comprise a bivariate distribution.

39. The method of claim 38, wherein said bivariate distribution is a parametric distribution.

25 40. The method of claim 38, wherein said parametric distribution is a bivariate normal distribution.

41. The method of claim 40, wherein said bivariate normal distribution and said system parameter further comprises a multiplicative error term consisting of a standard deviation with respect to a mean of signal 5 x ($\sigma_{\varepsilon x}$), a standard deviation with respect to a mean of signal y ($\sigma_{\varepsilon y}$) and a correlation between signals x and y (ρ_{ε}), and an additive error term consisting of a standard deviation with respect to a mean of signal x ($\sigma_{\delta x}$), a standard deviation with respect to a mean of signal 10 y ($\sigma_{\delta y}$) and a correlation between signals x and y (ρ_{δ}).

42. The method of claim 39, wherein said parametric distribution is a t-distribution.

43. The method of claim 39, wherein said parametric distribution is a bivariate gamma 15 distribution.

44. The method of claim 34, wherein said mean signal pair per analyte and system parameter are determined at the same time.

45. The method of claim 34, wherein said 20 system parameter is determined before said mean signal pair per analyte is determined.

46. The method of claim 45, wherein said predetermined system parameter is used to determine said mean signal pair per analyte.

47. The method of claim 34, wherein said enhanced values for said probability likelihood of said observed signals are produced one or more times until said mean signal pair per analyte and said system 5 parameter converge.

48. The method of claim 34, wherein determining said mean signal pair per analyte and said system parameter further comprises a non-linear optimization algorithm.

10 49. The method of claim 48, wherein said optimization algorithm is selected from the group consisting of Gradient Descent, Newton-Raphson and Simulated Annealing.

15 50. The method of claim 34, further comprising identifying significantly unequal mean signal pairs per analyte by a statistical difference indicator.

51. The method of claim 50, wherein said difference indicator further comprises a generalized likelihood ratio test statistic (λ).

20 52. A method of determining relative amounts of an analyte between samples, comprising:

(a) obtaining observed signals x and y for an analyte within two or more sample pairs;

(b) providing a mean signal pair per analyte

25 (μ) and a system parameter (β) for each sample pair;

(c) computing a probability likelihood of said observed signals, said observed signals being related to said mean signal by an additive error (δ) and a

multiplicative error (ε), where said system parameter specifies the properties of said additive error and said multiplicative error, and

(d) selecting a mean signal μ and a system parameter (β) that provides a maximum probability likelihood of occurrence given said observed signals.

53. The method of claim 52, wherein said additive and multiplicative errors are independent with respect to each other.

10 54. The method of claim 52, wherein said observed signals and said mean signal pair per analyte within said sample pairs further comprise the relationship:

$$x_{ij} = \mu_{xi} + \mu_{xi}\varepsilon_{xij} + \delta_{xij}, \text{ and}$$
$$15 \quad y_{ij} = \mu_{yi} + \mu_{yi}\varepsilon_{yij} + \delta_{yij}$$

where each measurement j equals 1 through M and each analyte i equals 1 through N ; where x_{ij} and y_{ij} are the observed signals, and where μ_{xi} and μ_{yi} are the mean signals.

20 55. The method of claim 52, wherein said additive and multiplicative errors further comprise a bivariate distribution.

56. The method of claim 55, wherein said bivariate distribution is a parametric distribution.

57. The method of claim 56, wherein said parametric distribution is a bivariate normal distribution.

58. The method of claim 57, wherein said 5 bivariate normal distribution and said system parameter further comprise a multiplicative error term consisting of a standard deviation with respect to a mean of signal x ($\sigma_{\varepsilon x}$), a standard deviation with respect to a mean of signal y ($\sigma_{\varepsilon y}$) and a correlation between signals x and y 10 (ρ_{ε}), and an additive error term consisting of a standard deviation with respect to a mean of signal x ($\sigma_{\delta x}$), a standard deviation with respect to a mean of signal y ($\sigma_{\delta y}$) and a correlation between signals x and y (ρ_{δ}).

59. The method of claim 56, wherein said 15 parametric distribution is a t-distribution.

60. The method of claim 56, wherein said mean signal pair per analyte and system parameter are determined at the same time.

20 61. The method of claim 52, wherein said system parameter is determined before said mean signal pair per analyte is determined.

62. The method of claim 61, wherein said predetermined system parameter is used to determine said 25 mean signal pair per analyte.

63. The method of claim 52, further comprising computing said probability likelihood of said observed

signals one or more times until said mean signal pair per analyte and said system parameter converge.

64. The method of claim 52, wherein said mean signal pair per analyte and said system parameter are determined by a method selected from the group consisting of maximum likelihood estimation (MLE), Quasi-Maximum Likelihood and Generalized Method of Moments.

65. The method of claim 52, wherein selecting said mean signal pair per analyte and said system 10 parameter further comprises a non-linear optimization algorithm.

66. The method of claim 65, wherein said optimization algorithm is selected from the group consisting of Gradient Descent, Newton-Raphson and
15 Simulated Annealing.

67. The method of claim 52, further comprising identifying said mean signal pair per analyte that are significantly unequal using a difference indicator.

68. The method of claim 67, wherein said
20 difference indicator further comprises a generalized
likelihood ratio test statistic (λ).

69. The method of claim 67, further comprising
selecting two or more mean signal pairs per analyte
having a difference indicator greater than that
25 corresponding to a false positive error rate.

70. The method of claim 52, wherein said analyte is a nucleic acid or polypeptide.

71. A method of determining relative amounts of analytes between samples, comprising:

- 5 (a) obtaining observed signals x and y for a plurality of immobilized analytes within two or more sample pairs;
- 10 (b) determining a mean signal pair per analyte (μ) and a system parameter (β) for each sample pair that provides a maximum probability likelihood of occurrence given said observed signals, said observed signals being related to said mean signal by an additive error (δ) and a multiplicative error (ε), where said system parameter specifies the properties of said additive error and said
- 15 multiplicative error, and
- (c) identifying one or more mean signal pairs per analyte that is significantly unequal.

72. The method of claim 71, wherein said additive and multiplicative errors are independent with respect to each other.

73. The method of claim 71, wherein said observed signals and said mean signal pair per analyte within said sample pairs further comprise the relationship:

$$25 \quad x_{ij} = \mu_{xi} + \mu_{xi}\varepsilon_{xij} + \delta_{xij}, \text{ and}$$
$$y_{ij} = \mu_{yi} + \mu_{yi}\varepsilon_{yij} + \delta_{yij}$$

where each measurement j equals 1 through M and each analyte i equals 1 through N ; where x_{ij} and y_{ij} are the

observed signals, and where μ_{xi} and μ_{yi} are the mean signals.

74. The method of claim 71, wherein said one or more mean signal pairs per analyte are identified as
5 significantly unequal by using a difference indicator.

75. The method of claim 74, wherein said difference indicator further comprises a generalized likelihood ratio test statistic (λ).

76. The method of claim 74, further comprising
10 selecting two or more mean signal pairs per analyte having a difference indicator greater than that corresponding to a false positive error rate.

77. The method of claim 71, wherein said analyte is a nucleic acid or polypeptide.

15 78. The method of claim 71, wherein said plurality of analytes further comprises about 1,000 or more different analytes.

79. The method of claim 71, wherein said plurality of analytes further comprises about 10,000 or
20 more different analytes.

80. The method of claim 71, wherein said plurality of analytes further comprises about 30,000 or more different analytes.

81. The method of claim 71, further comprising
25 analytes mobilized on a microarray.

82. The method of claim 71, further comprising the steps of:

(a) obtaining one or more reference signals, and

5 (b) determining a mean signal pair (μ) and a system parameter (β) for a sample pair comprising said observed signal x or y and said reference signal that provides a maximum probability likelihood of occurrence given said reference and observed signals, said reference and observed signals being related to said mean signal by an additive error (δ) and a multiplicative error (ε), 10 wherein said system parameter specifies the properties of said additive error and said multiplicative error.

83. A method of determining relative amounts 15 of an analyte between samples, comprising:

(a) obtaining a reference signal;

(b) obtaining observed signals x and y for an analyte within two or more sample pairs;

(c) determining system parameters (β_1, β_2) for a 20 sample pair comprising said observed signals x or y and said reference signal that provide a probability likelihood of said occurrence given said observed and reference signals, said observed and reference signals being related to said mean signal by an additive error 25 (δ) and a multiplicative error (ε), where said system parameter specifies the properties of said additive error and said multiplicative error;

(d) determining mean signal pairs (μ_1, μ_2) for 30 said sample pair comprising maximizing a product of terms for said probability likelihood of said sample pair of observed signals x or y and said reference signal for said analyte, and

(e) selecting a mean signal μ_x or μ_y that provides a maximum probability likelihood of occurrence given said observed signals and system parameters β_1 and β_2 .

5 84. The method of claim 83, wherein said mean signal pairs (μ_1 , μ_2) are determined using β_1 and β_2 obtained from step (c).

85. A method of determining relative amounts of an analyte between samples, comprising:

10 (a) measuring observed signals x, y and z for an analyte within two or more sample sets, and
(b) determining a mean signal set per analyte (μ) and a system parameter (β) for each sample set that produce enhanced values for a probability likelihood for
15 said observed signals, said observed signals being related to mean signals by an additive error (δ) and a multiplicative error (ε).